



Virology & Laboratory Diagnosis of Dengue

Mohammad Hassan Pouriayevali, Ph.D.

Department of Arboviruses and Viral Hemorrhagic Fevers, Pasteur Institute of Iran
(National reference laboratory)

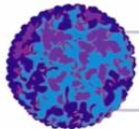
Overview

- ❖ **Virology**
- ❖ **Importance of Diagnosis**
- ❖ **Diagnostic methods**
- ❖ **Interpretation**

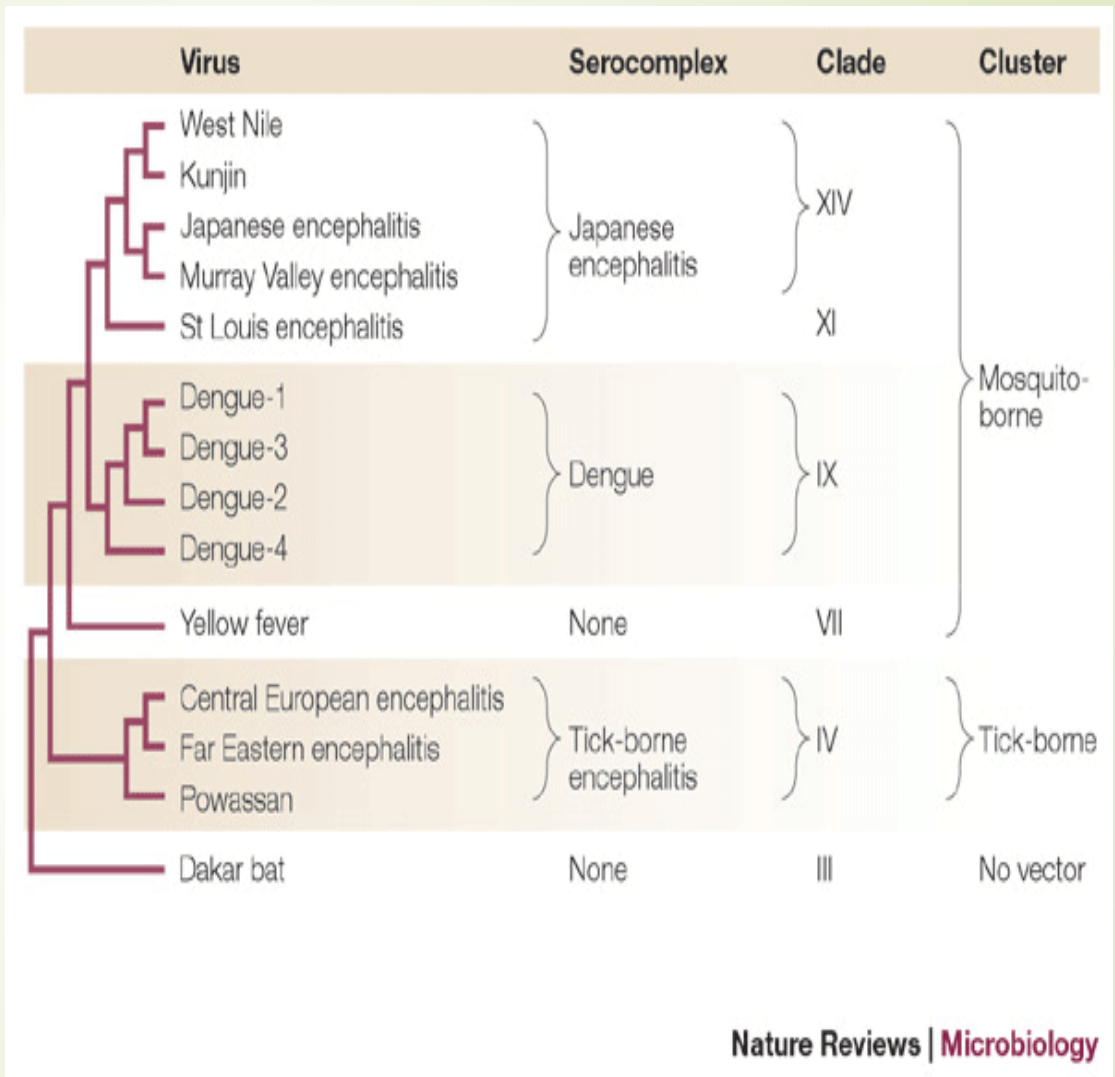
Dengue viruses

3

- Family **Flaviviridae**
- Genus **Flavivirus.**

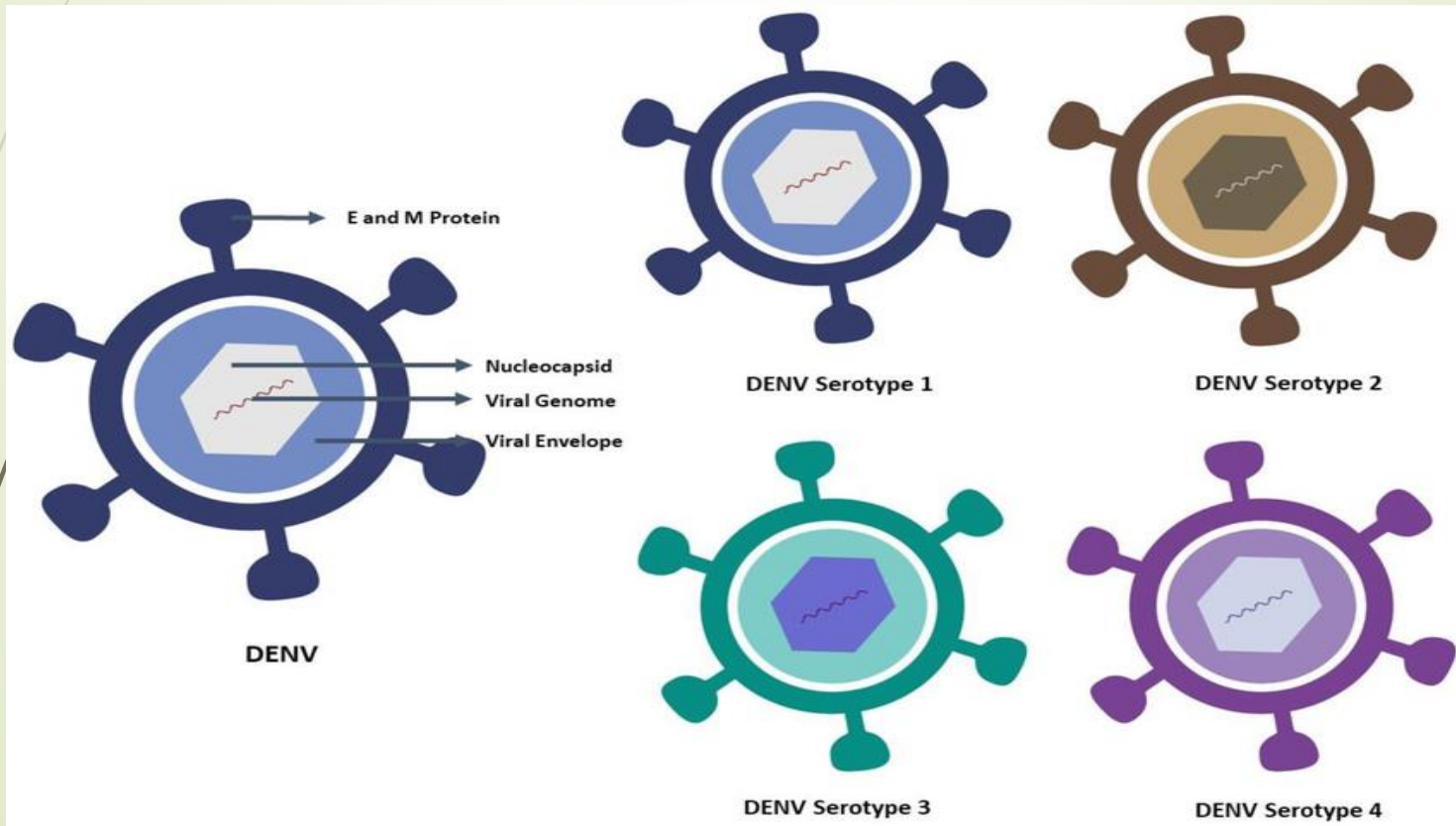


Dengue



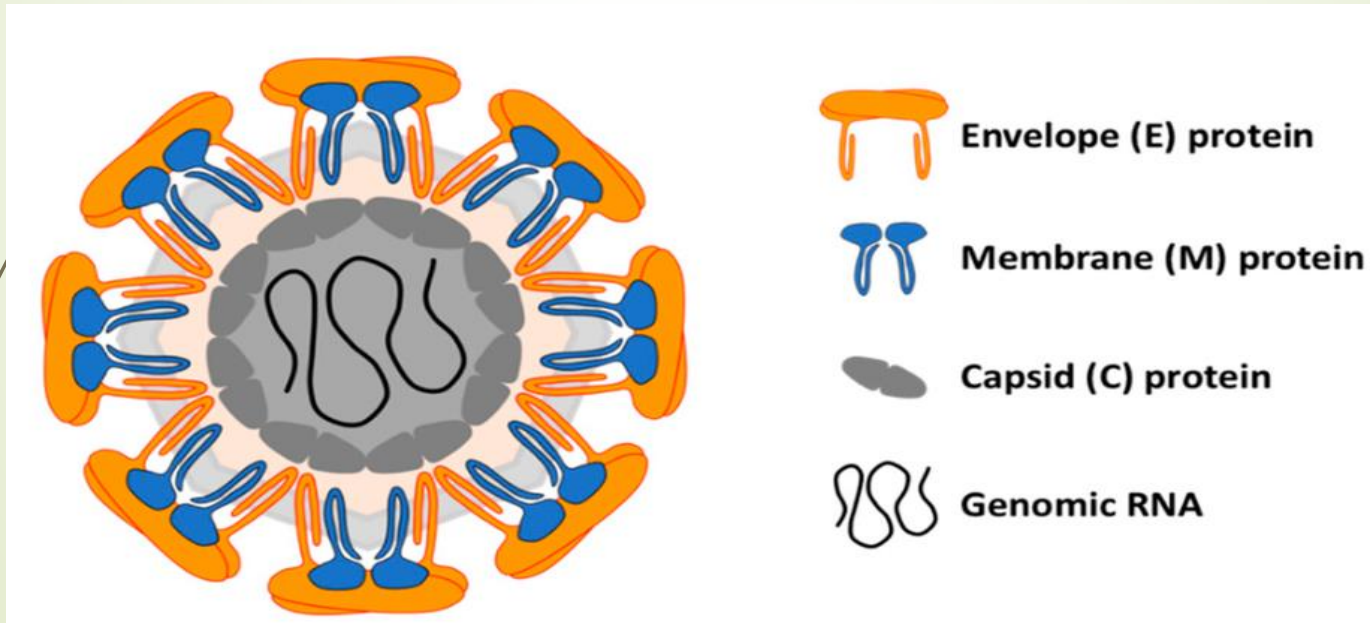
DENV serotypes

Four DENV serotypes can cause a spectrum of illness ranging from asymptomatic dengue infection to dengue fever (DF) to dengue hemorrhagic fever (DHF) to dengue shock syndrome (DSS)



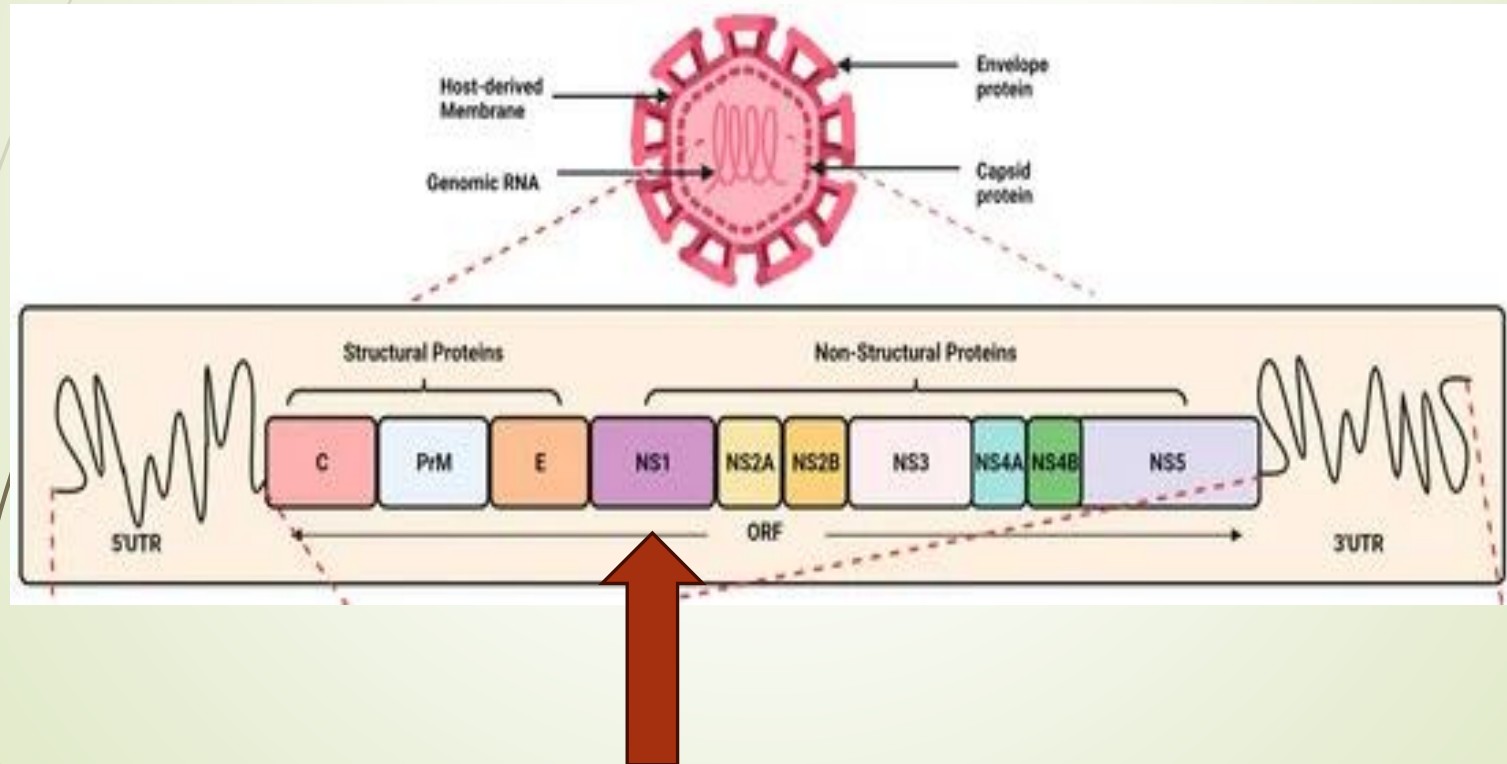
Morphological Properties

- A spherical enveloped virus (about 50 nm in diameter) with single-protein capsid, two membrane proteins, and single-stranded positive-sense ribonucleic acid (RNA) genome.



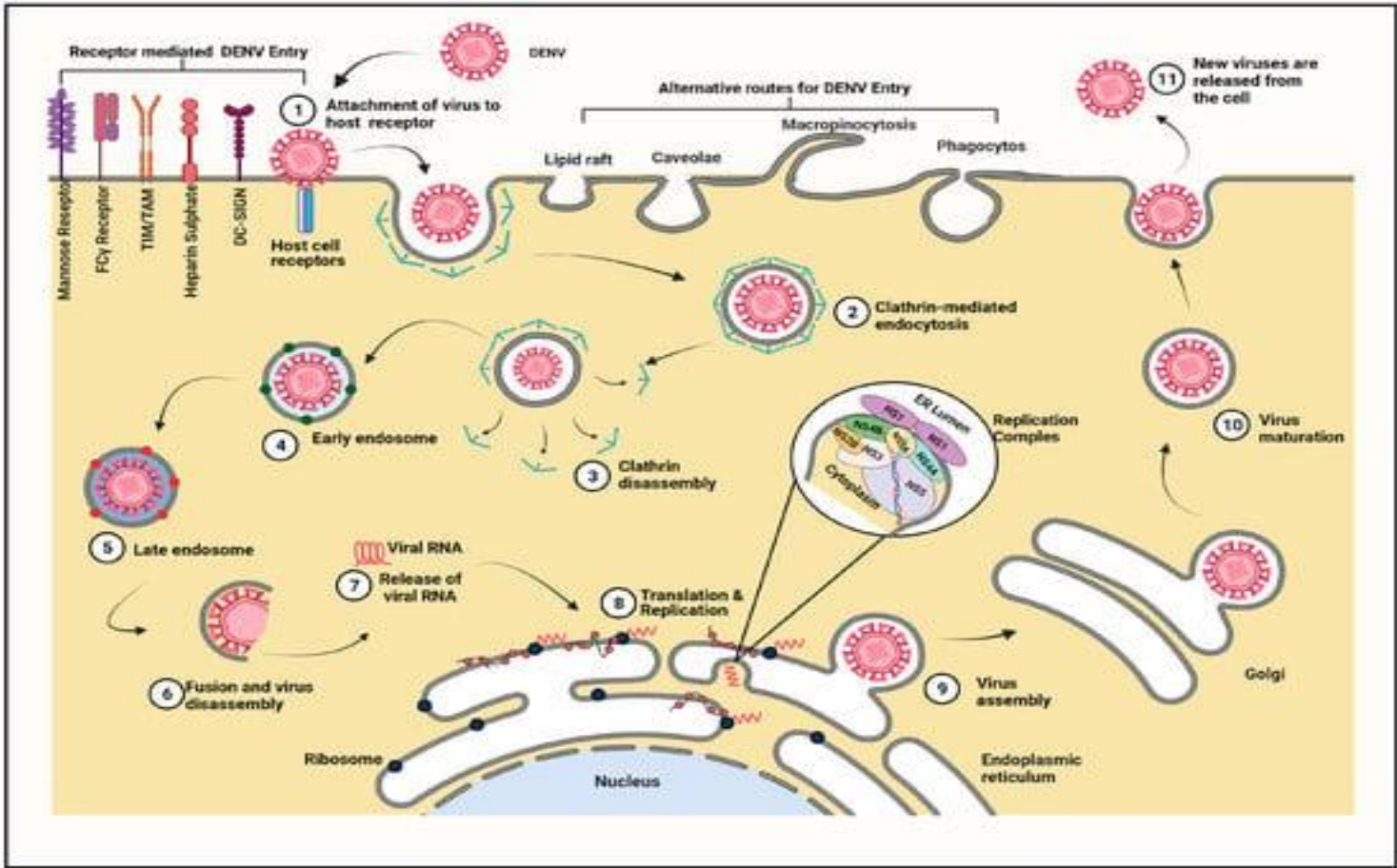
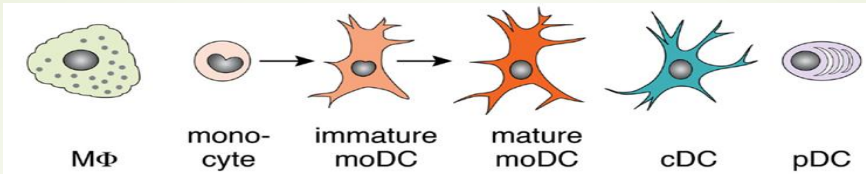
Genomic Properties

The Dengue genome is ~11 kb in length which is translated single complete polyprotein, with RNA helicase and RNA-dependent RNA polymerase (RdRp).



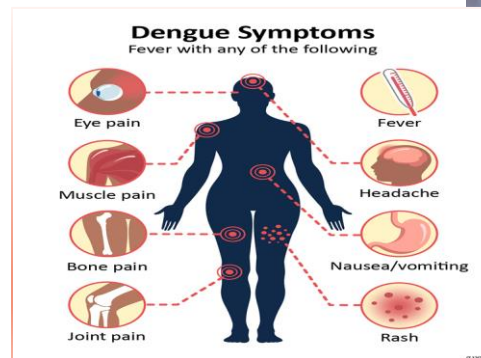
Viral Life Cycle

7



Importance of Diagnosis

- Surveillance activities
- Outbreak control
- Clinical management
- Differential diagnostic



Differential diagnostic

BACTERIAL

- Scarlet fever (Group A Strep)
- Meningococemia
- Typhoid fever
- Other bacterial septicemia
- Leptospirosis
- Rickettsial diseases

VIRAL

- Influenza
- Enterovirus
- Measles
- Rubella
- **CCHF**
- West Nile Virus
- Acute Retroviral Syndrome (HIV)
- **Chikungunya**
- Other alphaviruses
- Epstein-Barr virus
- Parvovirus
- Roseola virus

OTHER

- Malaria
- Post-infectious arthritis
- Rheumatologic diseases

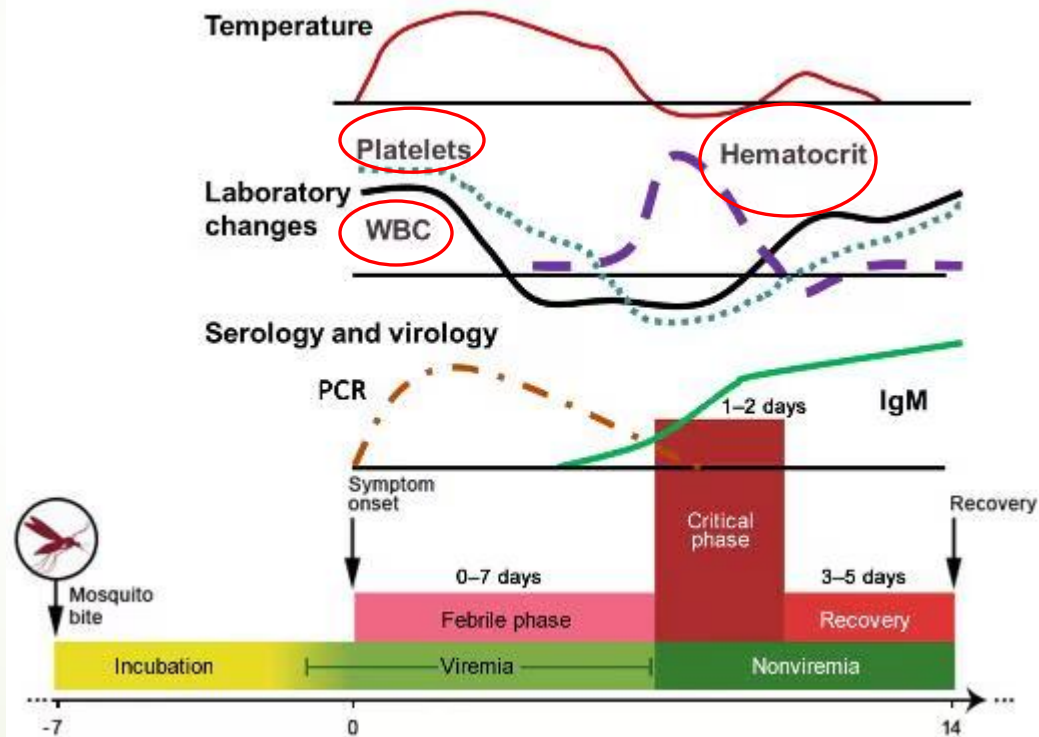
Importance of Comprehensive Assessment

- Accurate diagnosis relies on integrating laboratory findings with:
 - **Patient history:** especially travel to endemic areas
 - **Clinical symptoms** (e.g. fever, headache, arthralgia, myalgia, rash, nausea,)
 - **Routine clinical laboratory.**(WBC, HCT, Platelets,...)

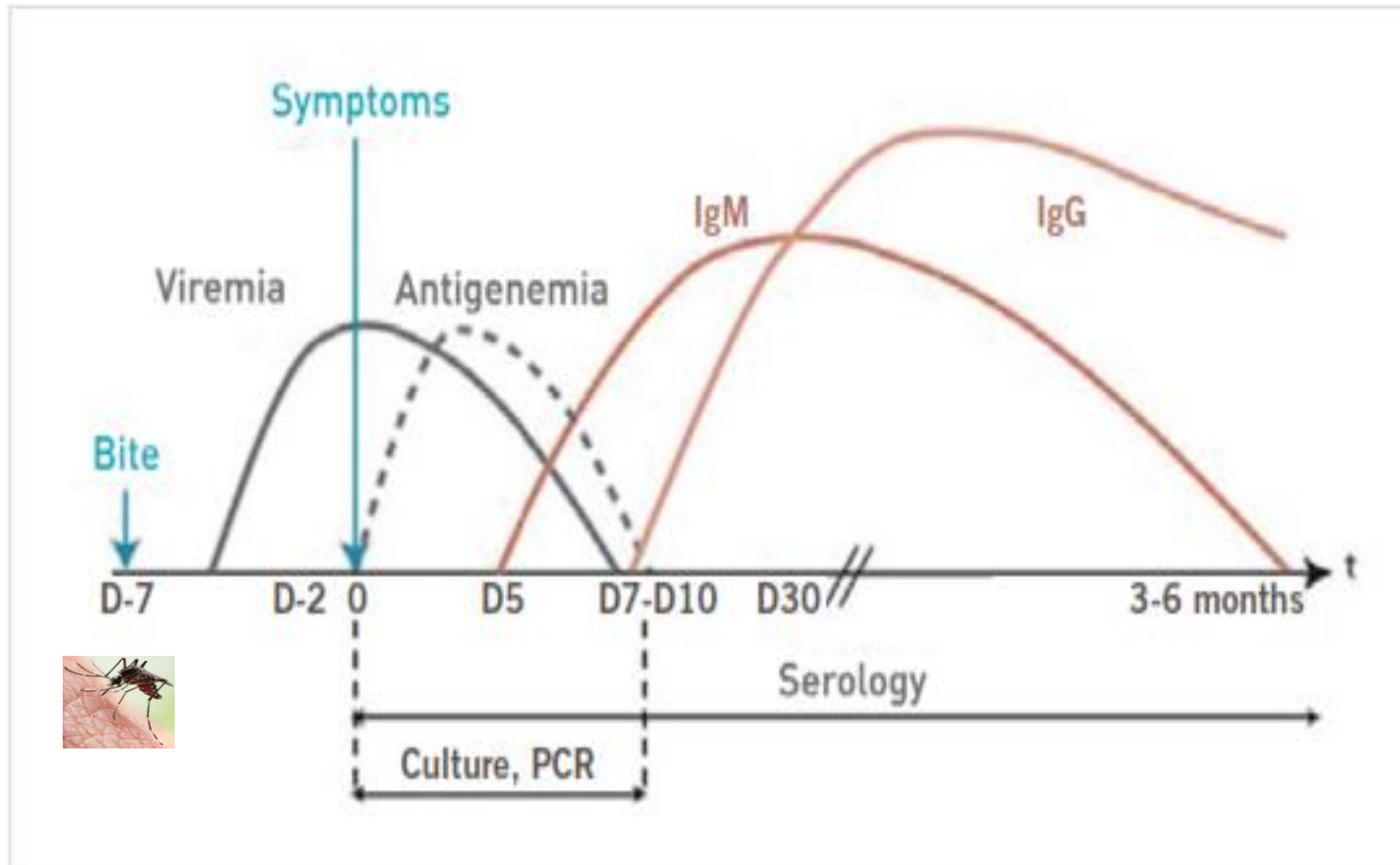
Routine laboratory tests

Laboratory findings	Chikungunya	Dengue	Zika
Anaemia	+	-	-
Leucopenia	++	+++	-/+
Neutropenia	+	+++	-
Lymphocytopenia	+++	++	-/+
Thrombocytopenia	+	+++	-/+*
Increased CRP	++	+++	-
Increased ALT	++	+++	-

+++ (very common) ++ (frequently observed), + (sometimes observed), - (no typical)

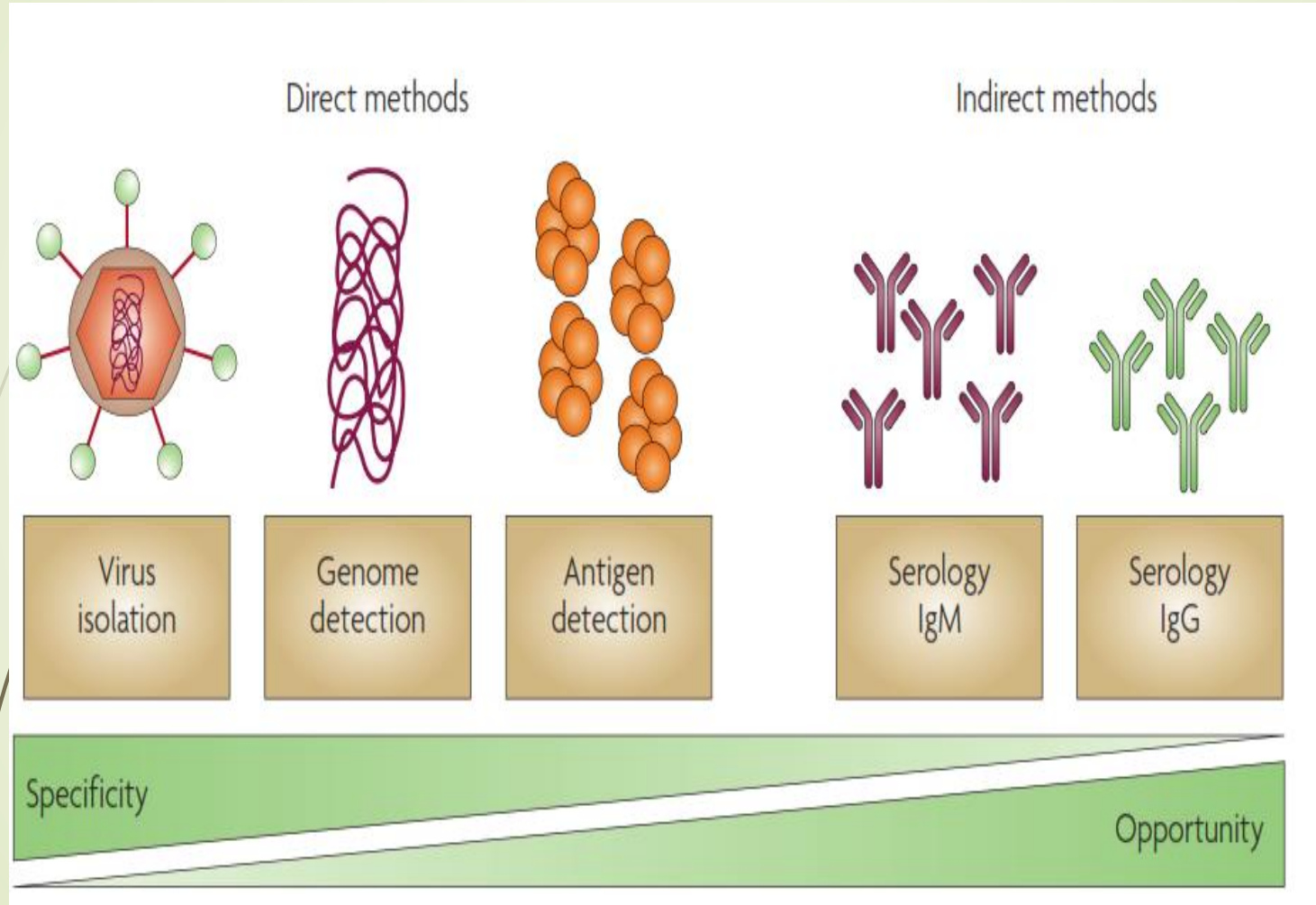


Dengue Virus infection Kinetics



Diagnostic methods

13



Kinetics of Infection and Antibodies Response

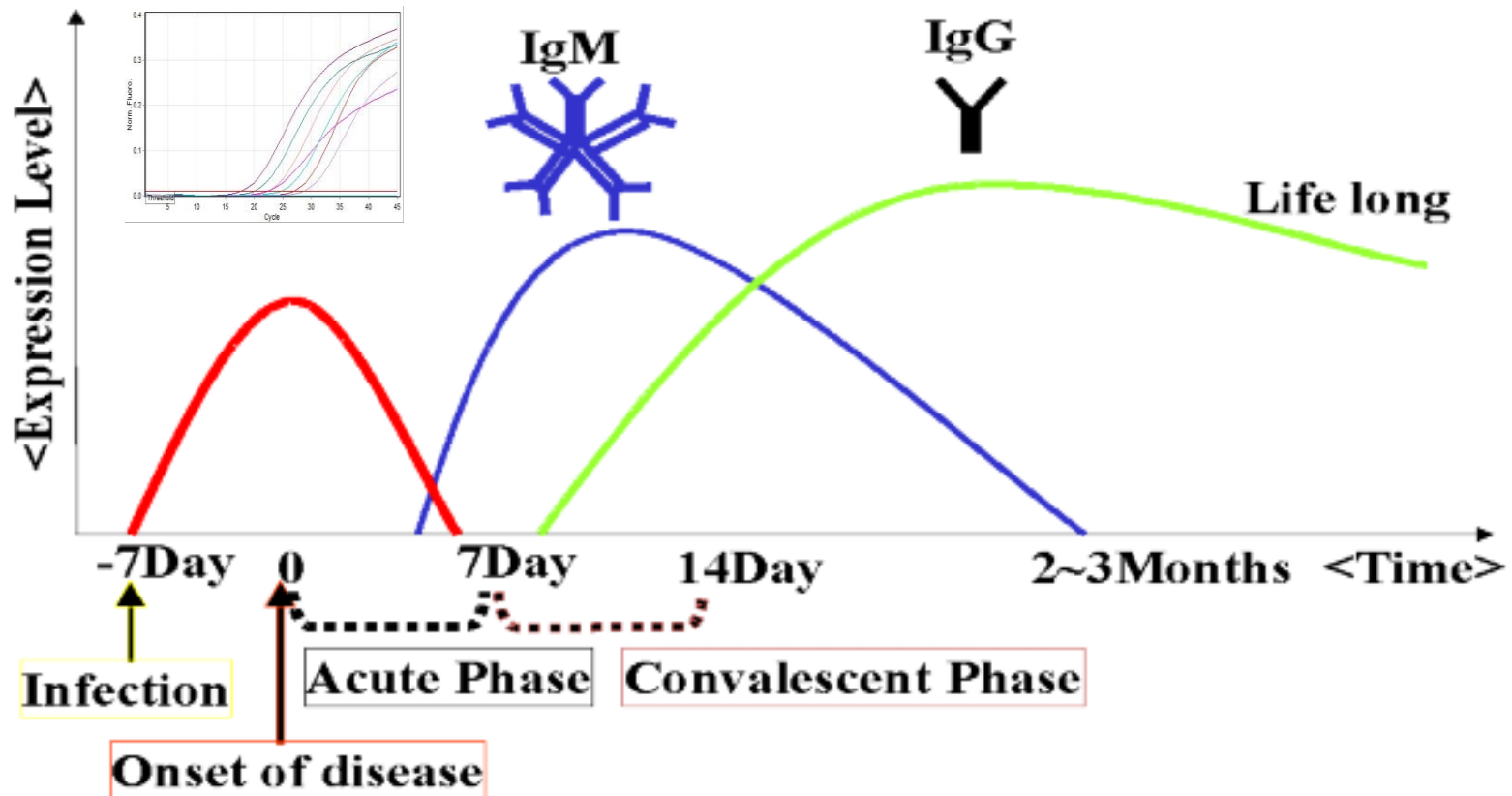
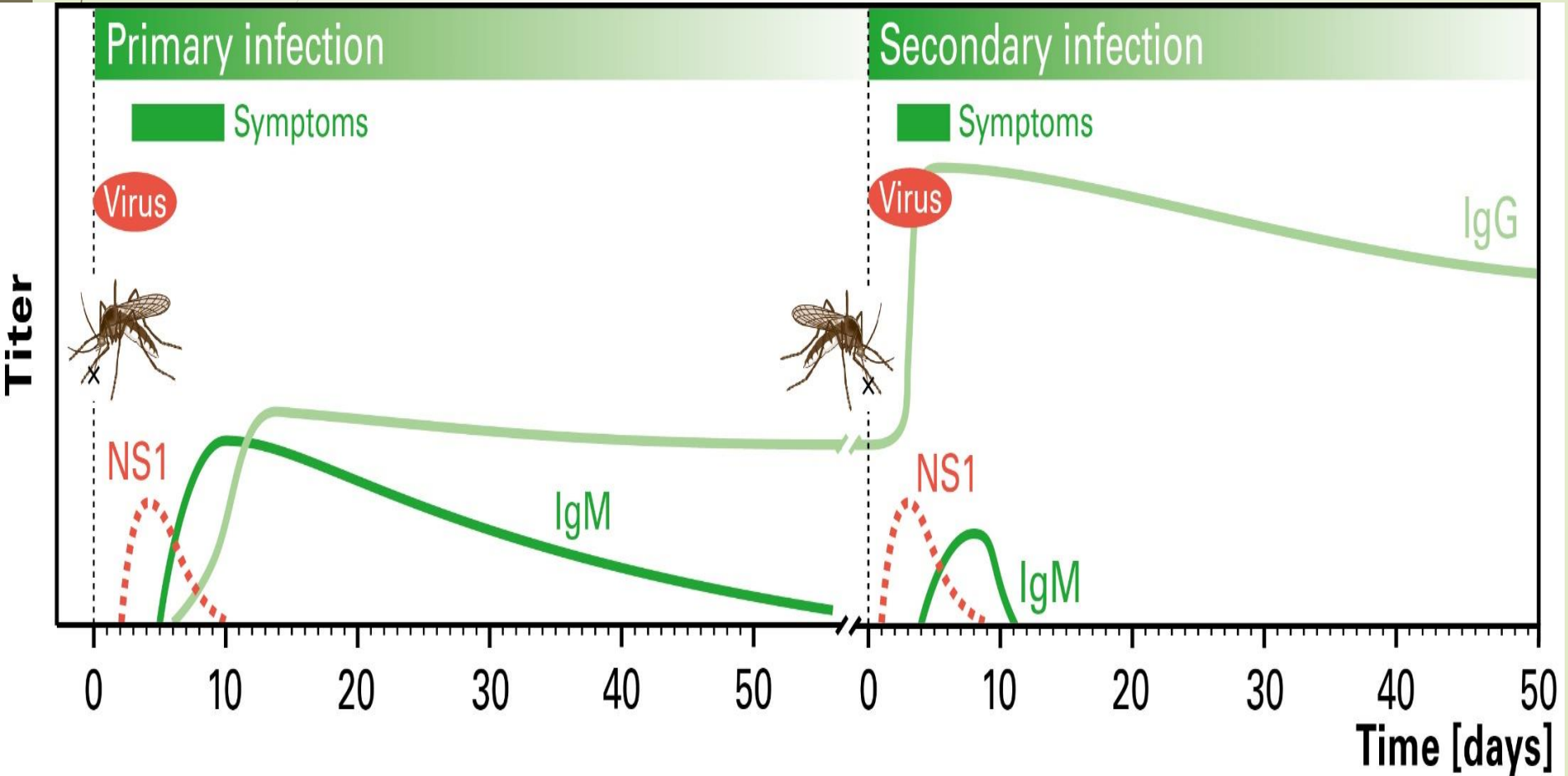


Figure 3: Kinetics of infection and antibodies response

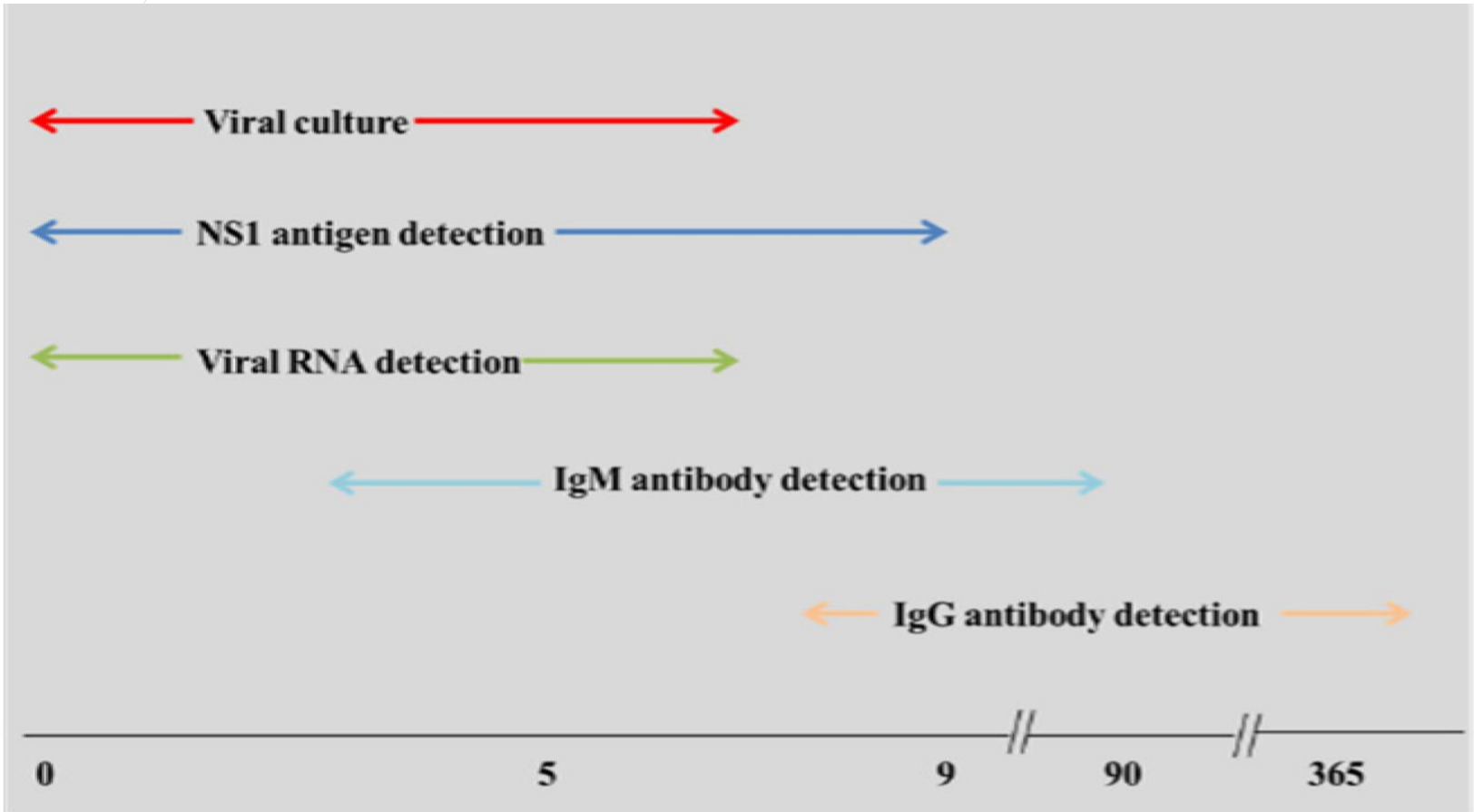
Primary vs Secondary infections

15



Combining different diagnostic methods

16



Sampling strategy

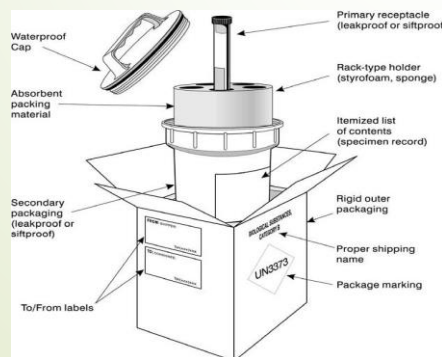
17

Sample Type: **Serum**

Two sets of samples are required for each DENV suspected case:

1: Acute phase sample (<7 days after onset of symptoms):
Real Time PCR& ELISA(IgM + IgG)

2: Convalescent phase sample (2 weeks after acute sample):
ELISA (IgM + IgG)



Interpretation

18

- Dengue virus infection is confirmed
 - RT-PCR or real-time RT-PCR are positive
 - Positive NS1 test

In paired serum samples

- IgM seroconversion
- IgG seroconversion
- Four-fold rise in IgG titer

- A presumptive dengue infection:
 - Positive IgM in a single serum sample
 - Positive IgG in a single serum sample

Single Specimen

	Laboratory tests				Interpretation
	NS1	Real time RT-PCR #	IgM	IgG	
Single specimen	+	+	+	+	DENV Infection
	+	+	+	-	DENV Infection
	+	+	-	+	DENV Infection
	+	+	-	-	DENV Infection
	+	-	-	-	DENV Infection
	+	unknown	unknown	unknown	DENV Infection
	unknown	+	unknown	unknown	DENV Infection
	-	+	-	-	DENV Infection
	+	-	+	+	DENV Infection
	+	-	+	-	DENV Infection
	+	-	-	+	DENV Infection
	-	+	+	+	DENV Infection
	-	+	+	-	DENV Infection
	-	+	-	+	DENV Infection
	-	-	+	+	Presumptive DENV Infection*
	-	-	+	-	Presumptive DENV Infection*
	-	-	-	+	Presumptive DENV Infection*
					Negative***

Paired Specimen

	Laboratory tests				Interpretation
	NS1	Real time RT-PCR #	IgM	IgG	
Paired specimen	-	-	-	-	DENV Infection
	-	-	+	-	
	-	-	-	-	DENV Infection
	-	-	+	+	
	-	-	-	-	DENV Infection
	-	-	-	+	
	-	-	+	-	DENV Infection
	-	-	+	+	
	-	-	+	+	DENV Infection
	-	-	+	-	
	-	-	+	+	DENV infection
	-	-	+	++**	
	-	-	+	+	Presumptive Past DENV or other falaviviruses Infection
	-	-	+	+	
	-	-	+	+	Presumptive Past DENV or other falaviviruses Infection
	-	-	-	+	
	-	-	-	-	Negative
	-	-	-	-	
	-	-	+	+	Inconclusive
	-	-	-/+	-	
-	-	+	+	Negative****	
-	-	-	-		

Rapid Diagnosis Assay

21

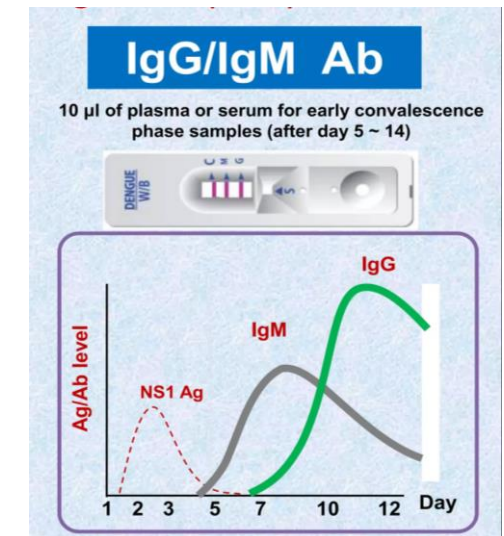
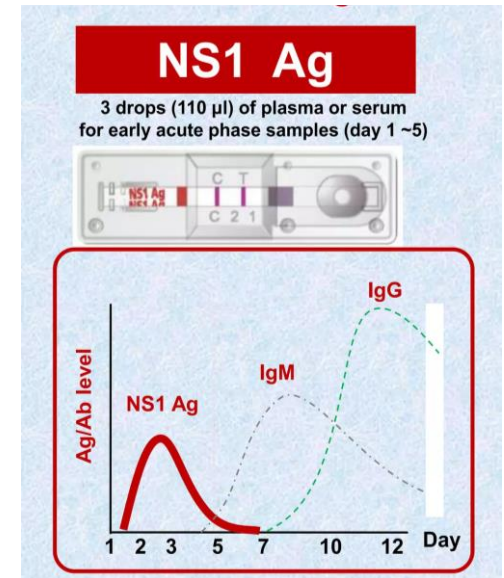
• Performance

- Sensitivity :

- 92.4% (Dengue NS1 Ag),
- 94.2% (Dengue IgG/IgM)

- Specificity :

- 98.4% (Dengue NS1 Ag),
- 96.4% (Dengue IgG/IgM)



22

THANK YOU

